Pedobiologia, Bd. 5, S. 146—152, 161—177 (1965)

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Observations on the breeding biology of Collembola (II)**)

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5.3. Fecundity

LUBBOCK (1873) reports that "the eggs are laid either singly, or in batches from fifty to a hundred", and to date the literature contains little more precise information. The most comprehensive data available are for *Sminthurus viridis*, where DAVIDSON (1934) records an average of 60 eggs per batch. HOLDAWAY (1927) and MACLAGAN (1932) have shown that the female of this species lays two egg batches at an interval of ten days. SOUTH (1959) records a mean of 53.5 for eleven batches of *Entomobrya multifasciata* (TULLBERG, 1871).

Just as observation on the sexual behaviour of Collembola is made difficult by the lack of a marked sexual dimorphism, so the design of experiments to determine the number of eggs laid is hindered. Four methods were attempted in the present work to obtain information on the number of eggs produced by an individual female. These methods are:

i. The estimation of the average size of the egg batch in mass cultures.
ii. The isolation of the gravid female until oviposition.
iii. The dissection of the gravid female.
iv. The setting up of a large number of small cultures containing two individuals, some 'pairs' thus being male and female. (The sexes of 'pairs' were determined on subsequent examination).

The methods of determining the numbers of eggs laid at each period of oviposition will be considered in turn:

i. The egg batch size.

Some information exists in the literature concerning the size of egg batches and this is summarised in Table 8. Some authors have indicated that their figures are means of a large number of egg batches, eg. BRITT (1951), but for the most part the number of egg batches is not indicated and no confidence limits are given. Probably the only figures that can be relied upon to be accurate estimates of the egg batch sizes are those of DAVIDSON (1934), BRITT (1951) and SOUTH (1959), the others being either too large or too small due to the methods used by the various workers.

At the beginning of the present work, several species were introduced into laboratory cultures in order to ascertain which species were most suitable for laboratory work. Several species produced eggs in mass culture, and a summary of the batch size in those species laying in this way is given in Table 2. Whilst some of the information is meagre because culturing of some of the species was discontinued, it is presented here since it is, in most cases, an improvement on the existing data.

The data shown for *Hypogastrura denticulata* agrees well with previous estimates of egg batch size in the genus, but in other genera the agreement is not so good. MILNE'S (1960) data for the genus *Onychiurus* are an underestimate for both soil dwelling (euedaphic) and litter dwelling (hemiedaphic) members. The data presented here show a

*) Now at: Department of Biology, Liverpool College of Technology (see p. 177).
**) Continuation of part I, Bd. 5, H. 1/2, 146—152.

Table 2 Egg batch sizes of Collembola reared in laboratory cultures.

Species	Number of batches	Mean number of eggs per batch	Standard deviation
Hypogastrura denticulata	145	30.2	31.3
Neanura muscorum	3	9.3	1.5
Onychiurus furcifer	54	14.3	6.5
Onychiurus procampatus	76	4.6	1.7
Onychiurus tricampatus	69	9.7	5.0
Onychiurus latus	41	15.1	5.6
Isotoma sensibilis	3	20.0	13.9
Isotoma viridis	8	21.5	9.9
Isotoma neglecta	40	34.9	39.2
Isotomurus palustris	12	30.3	21.5
Lepidocyrtus lanuginosus a.	21	16.4	17.0
b.	17	15.4	8.2

b. Insects from Moor House

a. Insects from Durham

significant difference ($P < 0.05$) between the sizes of the batches of the euedaphic Onychiuridae (*Onychiurus procampatus* and *Onychiurus tricampatus*) and the hemiedaphic species (*Onychiurus latus* and *Onychiurus furcifer*). The difference is emphasised most in a comparison of *Onychiurus latus* and *Onychiurus procampatus*, where both the adult females and the eggs are identical in size. Possibly the reduction in the size of the egg batch in euedaphic species is correlated with a reduced predation (or other mortality factor) in the more confined spaces between the soil particles, or possibly more batches may be laid. There is also a significant difference between the egg batch sizes of *Onychiurus procampatus* and *Onychiurus tricampatus*.

The size of egg batches in the Isotomidae appears to be generally larger than in the Entomobryidae in the present work, although DAVIS and HARRIS (1936) gave a high figure for *Pseudosinella petterseni* BÖRNER 1901 (Entomobryidae; see p. 167, Table 8).

ii. Isolation of the gravid female.

During the times of maximum egg laying in the field, presumably gravid adult females were collected and isolated in single cultures; it was hoped that some of these would prove to be fertilised females which would subsequently lay eggs. Fifty cultures of each of seven species were set up, and Table 3 gives the data obtained in this way.

Table 3 The number of eggs laid by isolated female Collembola.

Species	Total eggs	Number of cultures producing eggs	Mean number of eggs per female	Standard deviation
Hypogastrura denticulata	37	1	37.0	—
Isotoma viridis	51	1	51.0	—
Tomocerus minor	74	3	24.6	8.3
Lepidocyrtus lanuginosus	76	2	38.0	—
Dicyrtoma minuta	320	11	29.1	10.4
Dicyrtoma fusca	545	25	21.8	8.5

Note. In the first three species the data were obtained in May, in the last three species in October.

Except for the two species of *Dicyrtoma* the method proved to be unsatisfactory for determining the numbers of eggs laid by an individual female during a single period of egg laying. However, these results show that an adult female is capable of producing appreciably more eggs than the data for egg batch sizes would suggest. They also provide

information on the three species that did not lay eggs in batches in the cultures, namely *Tomocerus minor*, *Dicyrtoma minuta* and *Dicyrtoma fusca*.

iii. Dissection of the gravid female.

Of the four hundred insects isolated in the last experiment, half were used in other cultures after a period of three weeks, when it was assumed that no more eggs would be laid; the remaining two hundred were dissected. Some proved to be males, and in the majority of females the ovaries did not contain eggs. Only in the body cavities of females which had laid eggs were more eggs found, these individuals being dissected within 48 hours of ceasing oviposition.

The single *Hypogastrura denticulata* which laid 37 eggs contained 13 more, making a total of 50 eggs for the individual. In one female *Dicyrtoma minuta* which had laid 25 eggs, the right ovary was empty and the left contained 22 eggs; this gave a total of 47 eggs for the female. An individual which died after laying two eggs (not included in Table 3) was found to have 22 eggs in the right ovary and 24 eggs in the left, a total of 48 eggs. The other individuals of this species which were dissected were found to contain no more eggs. It seems probable that in this species and in *Dicyrtoma fusca* the eggs of one ovary are laid before the other, and that some individuals have already laid the contents of one ovary when collected from the field. Thus the data given in Table 3 are probably an underestimate of the numbers of eggs laid during a single period of oviposition.

Again the data in this section are meagre, and it is recognised that although eggs may be produced by the ovary this is no indication that the eggs would ever be laid. However, since batches of over 50 eggs have been recorded from *Hypogastrura denticulata* during the present work, and isolated females of *Dicyrtoma minuta* have laid up to 44 eggs, it is possible that all these eggs could be laid.

iv. Culturing 'pairs'.

The remaining individuals of the seven species of Collembola were isolated in pairs, but only one 'pair' produced eggs; this was a pair of *Onychiurus latus*, and 21 eggs, which subsequently proved to be fertile, were deposited in a single batch.

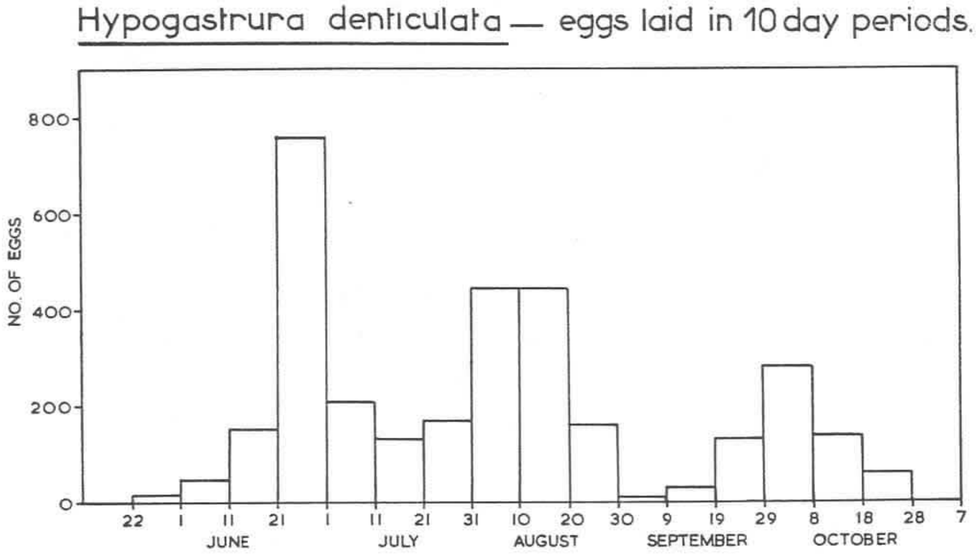
Holdaway (1927) and MacLagan (1932) have shown that two periods of oviposition occur in *Sminthurus viridis* at intervals of ten days, and Strebel (1932) reports that there are at least three batches of eggs produced in the lifetime of *Hypogastrura purpurescens*. Britt (1951) records that many female *Hypogastrura armata* produced two batches of eggs, and that one individual laid three batches and another four batches. Schaller (1953) reports three periods of egg laying in *Orchesella villosa*, and South (1959) records two females of *Entomobrya multifasciata* laying a total of eleven batches. On the basis of Lindemann's (1950) work, Handschin (1953) reports that in the genus *Orchesella* laying occurs at intervals after the fifth moult, "so that in the whole of an individuals's lifetime 60—80 eggs may be laid". Marshall and Kevan (1962) record four periods of oviposition in *Folsomia candida* (Willem 1902) and Sharma and Kevan (1963a) four periods in *Isotoma notabilis* Schäffer 1896.

It would appear then that the egg batch size and estimates of the number of eggs in the ovaries of a female at any given time would give an underestimate of the number of eggs produced in the lifetime of an average female, since each appears to lay more than once. Thus the data given previously may be regarded as minimum estimates of fecundity.

In two species, *Hypogastrura denticulata* and *Isotoma olivacea*, data are available from insects collected in the field before laying began. Histograms showing the numbers of eggs produced over different periods of the life of the insects in culture (Fig. 3) demonstrate that in *Hypogastrura denticulata* probably three periods of peak egg production occurred, and in *Isotoma olivacea* four such probable periods, were evident. No eggs were laid after 28 October in *Hypogastrura denticulata* and 15 July in *Isotoma olivacea*. Insects in both cultures were of maximum (adult) size, and most probably had not laid before being

introduced to the cultures; insects of the same species collected from the same habitat on previous dates did not lay until about the same time.

HANDSCHIN (1953) has recorded that *Orchesella* lays eggs before reaching a maximum size, and BRITT (1951) has made similar observations in *Hypogastrura armata*. MILNE (1960) records sexual maturity as being reached in the penultimate instar of *Onychiurus furcifer*,



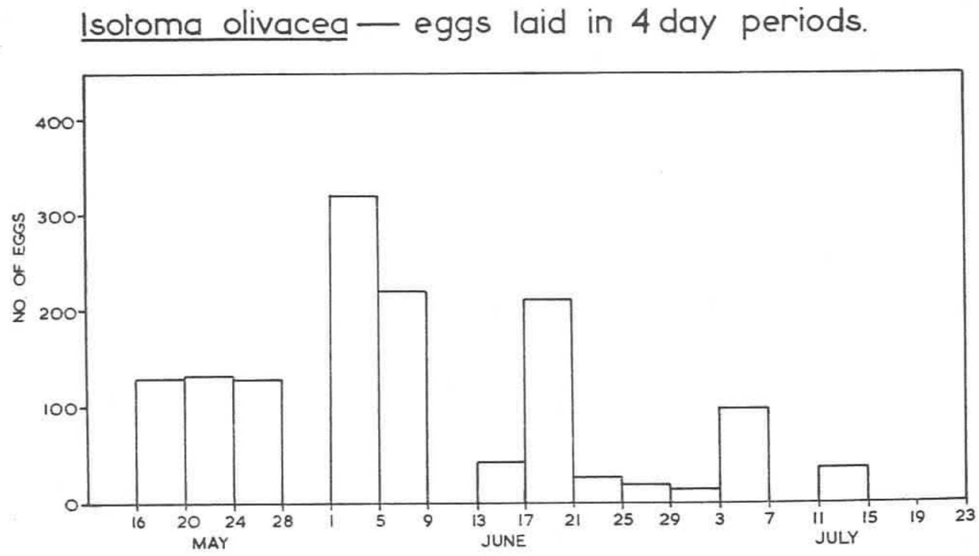


Fig. 3. Histograms showing the numbers of eggs laid in cultures by about 100 females of *Hypogastrura denticulata* over a period of five months and by about 50 females of *Isotoma olivacea* over a period of two and a half months.

Onychiurus latus, *Onychiurus procampatus* and *Tullbergia krausbaueri*, but since the data provided for the instar groupings are meagre, and do not agree with those obtained by the present writer for the same species (HALE, 1965); this information is regarded as unreliable. In the present work it has been shown that *Lepidocyrtus lanuginosus* reaches maturity before reaching maximum (adult) size. Fifth instar insects, some eight weeks old, laid eggs which were morphologically identical with those laid by adults, and these eggs developed and hatched over an identical period. Table 4 shows the egg batch size of individuals not having attained maximum size compared with that of adult *Lepidocyrtus lanuginosus*.

Qualitative observations suggested that eggs were produced by individual females directly after moulting in *Dicyrtoma minuta* and *Dicyrtoma fusca*, and in order to ascertain if this also occurred in members of the Arthropleona, a species which cultured easily and moulted frequently was sought; *Tullbergia krausbaueri* was found to fit these requirements. Cultures were set up in 'pairs', newly hatched first instars being used. These cultures proved to be much more successful than those of the other species previously recorded, and data were obtained from twenty three individuals. Table 5 shows in which instar the first eggs were produced, maximum size being reached in instar 4. It was found that once the female had begun to lay, it laid directly after moulting at successive moults. Although several individuals refrained from laying after moulting on some occasions, if laying was resumed this always occurred directly after another moult.

Table 4 Egg batch sizes at different ages in *Lepidocyrtus lanuginosus* in culture.

Age & instar	Number of eggs	Number of batches	Mean number of eggs per batch	Difference & S. E. of Difference	P
8 weeks (5th. instar) . .	57	10	5.7	8.5 ± 3.4	< 0.05
12 weeks (8th. instar) . .	71	5	14.2	22.1 ± 8.3	< 0.05
Adult	218	6	36.3		

Table 5 Egg production in the first ten instars of *Tullbergia krausbaueri*.

Instar	1	2	3	4	5	6	7	8	9	10
Number laying for first time	0	0	1	8	7	2	3	0	1	1

Note. Maximum size is reached in the fourth instar.

Table 6 shows the mean number of eggs produced during each instar by twenty-three females of *Tullbergia krausbaueri*. The maximum number of eggs is laid in instar 7 or 8, after which the number produced after each moult decreases. Thus, even after attaining maximum size in instar 4 the number of eggs produced after each moult increases until instar 7 or 8.

It is of interest to compare these data for *Tullbergia krausbaueri* with the average egg batch sizes at different stages in the cultures shown in Fig. 3. These data are given in Table 7. In both cases there is a tendency for the egg batch sizes to be reduced in older insects, as has been shown in *Tullbergia krausbaueri*.

The data given in Table 3 for average batch size are open to criticism on the following lines: it cannot always be assumed that eggs in one cluster are the produce of a single female; DAVIDSON (1934) has found clusters of up to 400 eggs in cultures of *Sminthurus viridis* and BRITT (1951) points out that large clusters of eggs in cultures of *Hypogastrura armata* were the product of more than one female. This has also been suggested by PACLT

Table 6 Numbers of eggs produced in each instar in *Tullbergia krausbaueri*.

Instar	Total eggs	Number of batches	Mean no. of eggs per batch		Standard deviation
3	2	1	2.00		—
4	24	9	2.66		1.20
5	61	16	3.81		1.60
6	62	10	6.20		2.85
7	86	12	7.16		3.12
8	58	8	7.25		2.55
9	61	9	6.78		3.30
10	47	7	6.71		3.44
11	62	10	6.20		3.48
12	56	9	6.22		3.00
13	45	8	5.63		1.98
14	8	3	2.66		1.21
15	10	3	3.33	Av. 3.55	2.60
16	14	3	4.66		1.53

Table 7 Variations in egg batch size with age in *Hypogastrura denticulata* and *Isotoma olivacea*.

Species	Mean number of eggs per batch and Standard Deviation			
	1st. peak 22. 6—21. 7.	2nd. peak 21. 7.—30. 8.	3rd. peak 30. 8.—28. 10.	4th. peak —
Hypogastrura denticulata	21.5	32.0	25.0	
S. D.	12.3	32.2	14.9	
	16. 5.—28. 5.	28. 5.—13. 6.	13. 6.—29. 6.	29. 6.—15. 7.
Isotoma olivacea	65.3	41.9	28.1	14.7
S. D.	46.7	45.5	35.2	5.6

(1956). In the present work clusters of up to 176 eggs have been found in *Isotoma olivacea* and up to 160 in *Hypogastrura denticulata*. On four occasions in this last species eggs in which the chorion had split were allowed to remain in the cultures, and newly laid eggs were subsequently found in the same cluster; this supports the observations of the previous workers. This possibility of error was reduced by removing eggs from the cultures daily. Underestimation of the average batch size would be brought about by the laying female being disturbed; in the opinion of the present writer this is much the more common occurrence, and thus the figures given in Table 2 are probably a slight underestimate; that they are not a gross overestimate is shown by comparison with Table 3 which gives an indication of the number of eggs an individual female is capable of laying during a single period of oviposition.

An estimate of the number of eggs laid by a female during life can be obtained by multiplying the average batch size by the number of laying periods. It is thought that such a figure gives an indication of the minimum number of eggs that an individual female is potentially capable of laying during life, and Table 8 summarises this information.

The fecundity of females in the field is difficult to estimate. Factors such as the incidence of fertilisation, mortality and temperature of the environment (which affects the number of moults and thus the number of egg-laying periods, at least in some species) would have to be taken into consideration in the estimation of an average number of eggs per female during life. The temperature (8 °C) at which most of the mass cultures were maintained lies just below the mean temperature experienced by laying females at Moor House during spring, and just above the mean temperatures to which autumn layers are

Table 8 Estimates of fecundity in Collembola.

Species	Authority	Temp. of culture in °C	Mean no. of eggs in batch	Probable no. of layings	Estimated no. of eggs laid in life
Hypogastrura manubrialis	RIPPER 1930	22	30	3	90
H. purpurescens	STREBEL 1932	—	20—30	3	60—90
H. denticulata	HALE	8	30.2 ± 2.6	3	90
H. armata	BRITT 1951	24?	28	3	84
Neanura muscorum	HALE	8	9.3 ± 0.9	3?	28?
—	MILNE 1960	12	1—2	—	—
Onychiurus furcifer	HALE	15	14.3 0.9	2	28
—	MILNE 1960	12	1—6	—	—
O. procampatus	HALE	15	4.6 ± 0.2	2	9
—	MILNE 1960	12	1—2	—	—
O. latus	HALE	8	15.1 ± 0.1	2?	30?
—	MILNE 1960	12	1—2	—	—
O. tricampatus	HALE	15	9.7 ± 0.6	2	19
Tullbergia krausbaueri	HALE	15	5.5 ± 0.3	10	55
—	MILNE 1960	12	1—2	—	—
Folsomia similis	SHARMA & KEVAN 1963b	24?	3—14	—	—
F. candida	MILNE 1960	12	9—36	—	—
Isotoma sensibilis	MARSHALL & KEVAN 1962	24	9.7	4	39
—	HALE	8	20.0 ± 8.0	3	60
I. notabilis	SHARMA & KEVAN 1963a	17	7—8	4	28—32
I. viridis	HALE	8	21.5 ± 3.5	3	65
—	MILNE 1960	12	27—54	—	—
I. olivacea	HALE	8	34.9 ± 6.1	3	105
Isotomurus palustris	HALE	8	30.3 ± 6.1	3	91
—	JAMES 1933	—	19	—	—
Entomobrya multifasciata	SOUTH 1961	17	53.5	5—6	300
Orchesella villosa	LINDEMANN 1950	—	4—8	10	60—80
Lepidocyrtus lanuginosus	HALE	8	15.5 ± 2.0	3	47
Pseudosinella petterseni	DAVIS & HARRIS 1936	25	c 45	—	—
—	SHARMA & KEVAN 1963c	24	6—15	—	—
P. alba	SHARMA & KEVAN 1963c	24	6—16	—	—
Tomocerus minor	HALE	8	24.7 ± 4.8	—	—
Sminthurides aquaticus	ANDERS 1959	—	c 16	—	—
Sminthurus viridis	DAVIDSON 1934	—	60	2	120
Dicyrtoma minuta	HALE	8	29.1 ± 3.2	2	58
D. fusca	HALE	8	21.8 ± 1.7	2	44

subject. The data presented here for 8 °C can probably be regarded as a minimum estimate under the field conditions prevailing at Moor House. The species maintained at 15 °C had the growth rate increased by a factor of 2.8 over those at 8 °C; that is to say that whereas the individuals of *Hypogastrura denticulata* had a period of 8.0 ± 0.4 days between moults at 15 °C this was increased to 22.6 ± 2.5 days at 8 °C (HALE, 1965). Thus whilst members of the genus *Onychiurus* could be expected to moult at least twice during the period in which eggs are laid at Moor House (May to October), thus probably producing two egg batches, *Tullbergia krausbaueri* would not moult on ten occasions as indicated in Table 8 during this period. Probably only in this last species, which would moult about six times, laying a mean of about 33 eggs during the period of egg laying, are the data given in Table 8 an overestimate for the situation in the field at Moor House.

5.4. Laying period in the field

In an attempt to determine at what time of the year Collembola laid under field conditions at Moor House, two separate cultures for each of sixteen species were set up at monthly intervals, over a period of one year (January to December 1960). Twenty individuals collected in the field were placed in each culture. The cultures were maintained at a constant temperature of 8 °C. Egg laying in the cultures within fourteen days of setting up is indicated in Table 9 by a cross (×) for each culture, under the month in which the Collembola were collected. From this data it is clear that most species lay in late spring and early summer, and that eggs are not laid during the winter. Cultures of species that were collected during the first four months of the year, and did not produce eggs within fourteen days of collection, laid at a later date, suggesting that oviposition is inhibited during the winter months at Moor House. It was subsequently found that by raising the temperature at which the cultures collected in mid-winter were kept, oviposition could be brought about at an earlier date than in those maintained at 8 °C. It would thus appear that low temperatures inhibit oviposition in the field.

In the two species of *Dicyrtoma* two egg laying periods occur, one in summer and one in autumn. These two periods coincide with two separate generations; other species in Table 9 probably had only one generation in the year. In *Lepidocyrtus lanuginosus* the majority of eggs is laid in autumn, but a few individuals survive the winter and lay in the following spring.

Table 9 Estimated dates of laying in the field at Moor House, 1960.

Species	Month J	F	M	A	M	J	J	A	S	O	N	D
Hypogastrura denticulata	—	—	—	—	×	××	×	—	—	—	—	—
Neanura muscorum	—	—	—	—	—	××	×	—	—	—	—	—
Onychiurus procampatus	—	—	—	—	—	××	××	××	×	—	×	—
Onychiurus tricampatus	—	—	—	—	××	××	××	××	××	××	×	—
Onychiurus latus	—	—	—	—	××	××	—	—	—	—	—	—
Tullbergia krausbaueri	—	—	—	—	××	××	××	××	××	×	—	—
Isotoma sensibilis	—	—	—	××	××	×	—	—	—	—	—	—
Isotoma viridis	—	—	—	×	××	×	—	—	—	—	—	—
Isotoma olivacea	—	—	—	××	××	×	—	—	—	—	—	—
Isotoma infuscata	—	—	—	×	××	—	—	—	—	—	—	—
Isotomurus palustris	—	—	—	××	××	×	—	—	—	—	—	—
Lepidocyrtus lanuginosus	—	—	—	—	×	—	—	—	—	××	×	—
Tomocerus minor	—	—	—	×	××	×	—	—	—	—	—	—
Dicyrtoma minuta	—	—	—	—	—	—	×	—	—	××	××	—
Dicyrtoma fusca	—	—	—	—	—	—	×	—	—	××	××	—

— no eggs laid; × laying in one culture; ×× laying in two cultures

5.5. Development

5.5.1. *Morphological changes*

Directly the egg is laid, it begins to enlarge due to the uptake of water from the atmosphere. In an atmosphere of 100 % R. H. the eggs reach their maximum size in a few hours; further enlargement does not take place until the growth of the embryo begins after about 20 % of the developmental period. In fertile eggs the first sign of development of the embryo is see in the splitting of the chorion. That it is the growth of the embryo that causes the chorion to split is demonstrated by the fact that in infertile eggs no split occurs, although initial enlargement by water uptake takes place. In most genera the chorion remains attached as two cap-shaped structures at opposite poles of the egg, but in members of the genus *Onychiurus* which were cultured, the chorion remained closely

attached to the next membrane (Fig. 4) which will be referred to in what follows as the serosal cuticle.

On the splitting of the chorion the egg loses its spherical shape and takes the form of a flattened spheroid, except in the Entomobryidae and the two members of the genus *Dicyrtoma* which were cultured. The two halves of the membrane remain attached to the serosal cuticle throughout the course of the development of the egg. In all the species concerned in this study the chorion was found to be unpigmented, transparent and without any consistent pattern or appended structures. SOUTH (1961), however, has found a species-characteristic pattern in the chorion caps of members of the genus *Entomobrya*. The second membrane was found in those cases examined to be patterned, and in some cases it bore appendages. In the case of the Isotomidae the pattern was made up of small protuberances, but there was no consistency between the eggs of different individuals of the same species.

All three members of the Entomobryidae cultured had numerous long hair-like appendages arising from the serosal cuticle. NICOLET (in LUBBOCK 1873) comments on these processes, which HANDSCHIN (1926) considers may serve as an anchorage underground.

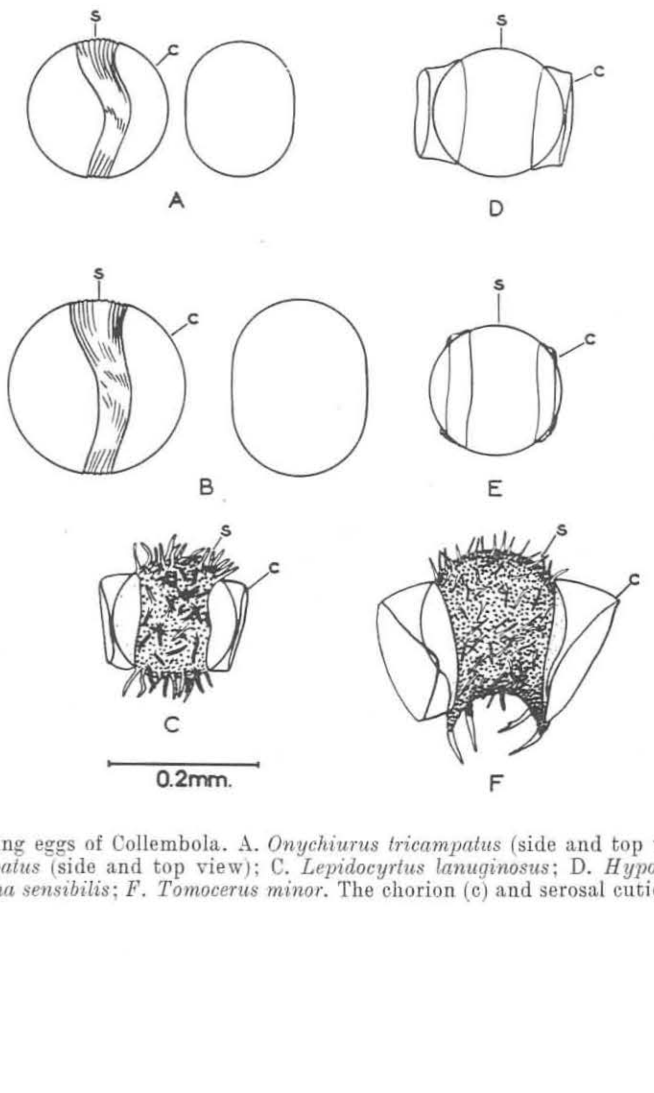


Fig. 4. Developing eggs of Collembola. A. *Onychiurus tricampatus* (side and top view); B. *Onychiurus procampatus* (side and top view); C. *Lepidocyrtus lanuginosus*; D. *Hypogastrura denticulata*; E. *Isotoma sensibilis*; F. *Tomocerus minor*. The chorion (c) and serosal cuticle (s) are indicated.

When the chorion first splits the serosal cuticle can be seen to be perfectly smooth in all three species cultured. The hairs grow out from the serosal cuticle over a period of six days at 8 °C, but no growth was observed beneath the chorion caps; only where the serosal cuticle was exposed to the air did hairs develop. In *Tomocerus minor* four large spines grew out from the serosal cuticle at the base of the egg, in addition to the hairs. UCHIDA and ABUKAWA (1956) referring to *Tomocerus minutus* TULLBERG 1876, record that "the chorion . . . forms a pair of lateral chambers each of which is soonly [*sic.*] provided with two curved spines". In fact there is no connection between the spines of the serosal cuticle and the chorion, as can be seen in Fig. 4. Each spine itself bears processes, and apically there is a long, claw-like region which curves under the egg; the length of each of the spines is approximately equivalent to the diameter of the egg. The fact that the processes are not produced by the egg until some days after it is laid may be correlated with a dispersal phase; Collembola laying this type of egg are surface and vegetation dwellers (members of GISIN's (1943) 'atmobios'), and the eggs will probably be more subject to dispersal from the position of laying than eggs of forms laying in the soil micro-caverns. The processes of the serosal cuticle, on their growing out, could act as anchors after dispersal of the eggs.

During further development, the eggs of the Isotomidae and Entomobryidae become coloured, after about 75 % of the developmental period has elapsed, apparently due to the formation of a pigment in the body of the embryo. However, eye-spots appear before the body colouration. The eggs of *Hypogastrura denticulata* remain colourless if kept in the dark, except for the eye spots which develop a dark pigment. If exposed to light all species which are pigmented in the adult stage tend to develop surface pigmentation and it was found that different coloured eggs could be produced dependent upon the time the eggs were exposed to light.

Table 10 Morphology of the eggs of Collembola.

Species	Colour of eggs on Laying	Colour of eggs on Hatching in dark	Diameter of eggs on Laying (mm)	Diameter of eggs on Swelling (mm)	Dimensions of eggs on chorion splitting (mm)	Dimensions of eggs on hatching (mm)
Hypogastrura denticulata	Colourless	Colourless	0.16	0.19	0.22	0.23
Neanura muscorum	Creamy-pink	—	0.28	0.34	—	—
Onychiurus furcifer	White	White	0.19	0.21	0.22 × 0.19	0.23 × 0.19
Onychiurus procampatus	White	White	0.23	0.26	0.27 × 0.22	0.28 × 0.22
Onychiurus tricampatus	White	White	0.17	0.20	0.20 × 0.17	0.21 × 0.17
Onychiurus latus	White	White	0.23	0.26	0.27 × 0.21	0.28 × 0.21
Tullbergia krausbaueri	White	White	0.08	0.09	0.12 × 0.10	0.12 × 0.10
Isotoma sensibilis	Orange-red	Orange-red	0.17	0.19	0.19 × 0.17	0.19 × 0.17
Isotoma viridis	Orange-red	Orange-red	0.20	—	—	—
Isotoma olivacea	Colourless	Pink	0.17	0.19	0.19 × 0.17	0.19 × 0.17
Isotoma infuscata	Colourless	Pink	0.17	0.19	0.19 × 0.17	0.19 × 0.17
Isotomurus	Yellow	Orange-pink	0.17	0.19	0.19 × 0.17	0.19 × 0.17
Lepidocyrtus lanuginosus	White	Pink	0.19	0.21	0.21 × 0.19	0.21 × 0.19
Tomocerus minor	Orange	Pink	0.21	0.23	0.23	0.23
Dicyrtoma minuta	White	Pink	0.28	—	—	—
Dicyrtoma fusca	White	Pink	0.28	—	—	—

Table 10 gives a summary of the morphology of the eggs of the species cultured during the present work. Measurements are for twenty or more eggs from at least two females in all cases.

5.5.2. *The Hatching period*

DAVIDSON (1934) has shown that the duration of development of *Sminthurus viridis* within the egg is dependent on both temperature and humidity. The present work is concerned with those Collembola living in the soil and litter layers where, according to THAMDRUP (1939), the relative humidity is greater than 90 % even during dry periods. No account has been taken of variations in humidity, except to maintain 100 % R. H. in culture jars containing eggs in the experiments involving different temperature conditions.

5.6. Egg development at constant temperature

Eggs laid in cultures were transferred on the day of laying to small culture tubes of the type described previously. These were then placed in constant temperature conditions, and a range between 2 °C and 23 °C was utilised. 2 °C, 4 °C, 7 °C, 12 °C, 16 °C and 23 °C were the temperatures most used, but others occasionally became available and were used when the opportunity arose. The following fourteen species were used in the experiments: *Hypogastrura denticulata*, *Onychiurus furcifer*, *Onychiurus procampatus*, *Onychiurus latus*, *Onychiurus tricampatus*, *Tullbergia krausbaueri*, *Isotoma viridis*, *Isotoma olivacea*, *Isotoma infuscata*, *Isotomurus palustris*, *Lepidocyrtus lanuginosus*, *Tomocerus minor*, *Dicyrtoma fusca* and *Dicyrtoma minuta*. Fig. 5 shows the relationship between temperature and the reciprocal of the development time (Velocity) in five of these species, one from each family of Collembola. The theoretical developmental zero for the five species concerned can be seen to be about 0 °C. For the other species concerned in this paper the developmental zero varies between 0 °C and 3 °C (see p. 172, Table 11). The development time was measured from the day of laying to the day when 50 % of the eggs in a given batch had hatched. Usually about twenty eggs were placed in a culture tube, but this was not always possible; occasionally where eggs were abundant in the cultures up to 50 eggs were used in each tube.

In cases where the velocity of development is proportional to the temperature, as in Fig. 5, if D is the development time in days, T the temperature in degrees centigrade, C the developmental zero in degrees centigrade and K a constant, the equation may be written:

$$D\,(T-C) \approx K$$

K, in degree-days, is termed the "thermal constant" (WIGGLESWORTH, 1953). Thermal constants for the species studied are shown in Table 11.

CHOUDHURI (1961) working on species of *Onychiurus* and SOUTH (1959) on species of *Entomobrya* found threshold temperatures of about 4 °C, below which no egg development occurred; in both cases specimens were collected from low lying ground (below 500ft ca. 150 m O. D.). In the present work development took place under experimental conditions at temperatures as low as 2 °C and theoretical developmental zero temperatures were found to lie between 0 °C and 3 °C. According to WIGGLESWORTH (1953) the temperature at which development ceases normally lies below the theoretical developmental zero. It thus appears possible that in the species involved in the present work there is a physiological mechanism allowing the eggs of high altitude forms to develop at temperatures below those at which it is possible for the same processes to be carried out at lower altitudes, in a relatively warmer climate.

The egg development period in different species appears to be unrelated to the food content (size) of the egg (compare Tables 10 and 11) or to any other obvious factor, such as availability to predators.

Table 11 Thermal constants at constant and fluctuating temperatures

Species	Developmental zero (°C)	Thermalconstant K at: Constant temp.	Fluctuating temp.
Hypogastrura denticulata . .	0	408	370
Onychiurus furcifer	3	276	—
Onychiurus latus	0	444	410
Onychiurus procampatus . .	3	355	—
Onychiurus tricampatus . . .	1	335	—
Tullbergia krausbaueri. . . .	0	510	—
Isotoma olivacea	0	195	180
Isotoma infuscata	1	134	—
Isotoma viridis	3	182	180
Isotomurus palustris	0	208	180
Lepidocyrtus lanuginosus . .	0	351†	340†
		328*	300*
Tomocerus minor	0	356	290
Dicyrtoma fusca	0	1260	—

† Eggs from Moor House * Eggs from Durham

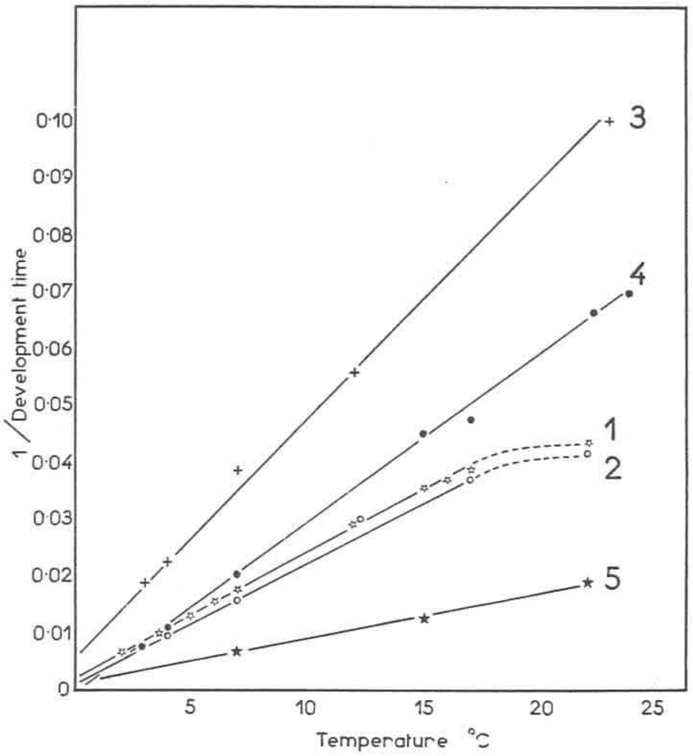


Fig. 5. The relationship between temperature and the reciprocal of the egg development time in days in five species of Collembola. 1. *Hypogastrura denticulata*. 2. *Onychiurus latus*. 3. *Isotoma olivacea*. 4. *Lepidocyrtus lanuginosus*. 5. *Dicyrtoma fusca*.

5.7. Egg development at fluctuating temperatures

Eggs of seven species of Collembola were placed in culture tubes in screened air, at the height of a Stevenson's screen, and subjected to the fluctuating outside air temperatures in Durham; the development time was observed as in the last section. Maximum and minimum temperatures were available at the screen, and median = (max + min) ÷ 2 temperatures were recorded daily over the development periods of all seven species. Thermal constants for fluctuating temperatures are shown compared with those for constant temperatures in Table 11. The figures for fluctuating temperatures are the median minus the developmental zero in degrees centigrade multiplied by the development time in days. The agreement between the thermal constants derived from the two different sources is such that an approximate prediction concerning hatching times in the field can be made, knowing the time of laying and the expected temperatures. The values of k for fluctuating temperatures were all lower than for fixed temperatures, which suggests a higher rate of development for the former.

5.8. Egg development in the field

In Table 9 it has been shown that the majority of species lay eggs in May and June at Moor House. From this information and the calculated thermal constants, estimates for the times of egg development in the field at Moor House have been made; median monthly screen temperatures over the period of ten years 1952—1961 have been used in these estimates, which are shown in Table 12.

Table 12 Estimates of egg development time in days in the field at Moor House, Westmorland (1840ft, ca. 560 m O. D.)

Species	Average of 10 years (1952—61). Devpt. time		1960 Devpt. time		1961 Devpt. time	
	Laying 1 May	Laying 1 June	Laying 1 May	Laying 1 June	Laying 1 May	Laying 1 June
Hypogastrura denticulata	50	40	45	37	53	43
Onychiurus procampatus	64	45	54	45	69	50
Onychiurus latus	54	44	47	40	57	47
Onychiurus tricampatus	44	37	40	32	48	38
Tullbergia krausbaueri	61	50	53	47	64	53
Isotoma viridis	39	28	33	22	42	29
Isotoma olivacea	27	20	24	17	30	21
Isotoma infuscata	22	16	19	14	24	17
Isotomurus palustris	29	22	25	18	32	23
Lepidocyrtus lanuginosus	43	35	38	30	46	36
Tomocerus minor	45	36	40	32	48	38
Dicyrtoma fusca	153	117	128	122	169	128
median temp. in °C	7.20	9.57	8.20	11.30	6.50	9.20

Note. Recent work suggests that *Dicyrtoma* spp. have two types of egg, summer eggs and over-wintering eggs; the data here are for the latter which apparently develop more slowly.

In order to test the accuracy of these estimates, two batches of eggs of *Hypogastrura denticulata* and two batches of *Onychiurus latus* were put out in the field at ground level at Moor House on 29 May, 1961. On 4 July, 1961 (after 36 days) 2 of 50 eggs of *Hypogastrura denticulata* had hatched and none of the eggs of *Onychiurus latus*. On 11 July, 1961 (after 43 days) all the eggs of *Hypogastrura denticulata* had hatched, and 50% (23) of the *Onychiurus latus* eggs had hatched. All the eggs had hatched by 17 July, 1961. These data give a period of about 40 days for the hatching of *Hypogastrura denticulata* in the field (calculated estimate for this in the field 43 days), and 43 days for *Onychiurus*

latus (calculated estimate 47 days). The faster rates are probably explained by the fact that cultures were placed at ground level where somewhat higher temperatures are to be expected than at screen height. It may be concluded that the estimates given in Table 12 for development times in the field approximate to the actual development times, and that the methods used for obtaining them are justified, in that these estimates approximate to actual development times measured in the field.

In Tabla 9 it was shown that *Lepidocyrtus lanuginosus* and the two species of *Dicyrtoma* laid the majority of their eggs in late autumn (October and November). The temperatures at Moor House at this time are so low that development would appear to be impossible until the following spring, and thus eggs must survive the winter. In order to test this hypothesis, twenty tubes, each containing a single egg were set in the ground at Moor House, for each of the three species concerned. The eggs were laid in culture on 8 November, 1959, and kept at 2 °C until placing in the field at Moor House on 23 November, 1959. The tubes were examined regularly throughout the winter, but it was not until 2 May, 1960 (175 days after laying) that any hatched. All the eggs of *Lepidocyrtus lanuginosus* hatched by 9 May, 1960 (between 175 and 182 days after laying). The two species of *Dicyrtoma* hatched between 23 May, 1960 and 6 June, 1960. First instar individuals were collected from the field populations for the first time in 1960 on the same dates that hatching was found in the cultures. This clearly shows that the eggs of these three species can readily survive the winter at Moor House.

Over-wintering eggs are at times subjected to temperatures well below freezing point, but this seems to have no effect on them other than to retard development. In Table 13 the results of freezing batches of eggs of *Hypogastrura denticulata* are shown; seven batches of about 30 eggs each were divided each into two. One part of each batch was maintained at 4.5 °C and the other was frozen for ten days at — 7 °C before being placed at the same temperature. The development time was increased by an average of 13.5 days in the frozen eggs. Eggs frozen for longer periods at — 7 °C were subject to a high mortality (over 80 %).

Table 13 The effect of freezing on the eggs of *Hypogastrura denticulata*.

Culture	Time in days for 50 % of the eggs frozen at — 7 °C. for 10 days and then kept at 4.5 °C to hatch	Time in days for 50 % of the eggs kept at 4.5 °C to hatch	Increase due to freezing (in days)
1	104	93	11
2	104	93	11
3	106	92	14
4	108	92	16
5	100	89	11
6	107	90	17
7	107	92	15
Average	105.1	91.6	13.5

6. The process of hatching

In those species of Collembola possessing pigmented eye spots, the onset of hatching can be forecast from the depth of the pigmentation. The embryo lies around the largest circumference of the egg, which at this time has the form of a flattened spheroid, in all except the members of the Entomobryidae and the two species of *Dicyrtoma*. The caps of the split chorion also lie on the largest circumference, opposite the head and the tip of the abdomen of the embryo.

MacLagan (1932) points out the lack of hatching spines or any similar structure in Collembola and indicates that eclosion is brought about by the movement of the embryo within the serosal cuticle. In the four species of *Onychiurus* cultured, the serosal cuticle was observed to be sculptured (Fig. 4, see p. 169), and the splitting of the serosal cuticle on eclosion occurred along the line of the sculpturing. In all cases except the two species of *Dicyrtoma*, the split occurred across the smallest diameter of the egg, in a plane at right angles to a line joining the two chorion caps; thus the splitting of the serosal cuticle on eclosion is in the same plane as the earlier splitting of the chorion. In the two species of *Dicyrtoma* the eggs were normally covered by faecal material, and eclosion occurred by individuals eating their way out of the top of the egg; no splitting of the serosal cuticle was observed.

In the species normally hatching by splitting, the serosal cuticle first splits opposite the gap between the embryonal head and the tip of the abdomen. The head and the antennae were forced through the transverse slit and the legs were then braced against the inside of the serosal cuticle; in this way the split was extended and the gap widened. Eclosion was often aided by the egg being anchored in some way to the substrate. In many cases this occurred by means of the projections of the serosal cuticle or by fungal hyphae which grew over the eggs. The sticking together of eggs in batches also aided eclosion, as the embryo was able to gain a purchase with its claws and drag the abdomen out of the serosal cuticle which remained attached to the rest of the egg batch. The furcula was frequently used in freeing the abdomen, especially where the egg had not become attached.

On freeing itself from the serosal cuticle the hatched individual usually retracted the furcula if it had been used in the process of hatching, and it was then immediately able to use it for jumping. Normally individuals took into their gut fungal mycelia and particles of the substrate within minutes of hatching.

7. Acknowledgements

The author wishes to thank Professor J. B. Cragg, Dr. J. C. Coulson and Dr. L. Davies for advice and encouragement during the course of this work which was carried out whilst holding a Durham Colleges Research Studentship. During the latter part of the study the facilities of the Zoology Department were made available by Professor David Barker and this is gratefully acknowledged. The Nature Conservancy allowed access to the Moor House National Nature Reserve.

8. Summary

The reproductive behaviour of Collembola collected from the Moor House National Nature Reserve, Westmorland, England (1840 ft, ca. 560 m O. D.) is described and the conclusions of Mayer (1957), that spermatophores are not produced by members of the Poduridae and the Onychiuridae, are supported. Spermatophores of *Tomocerus minor* were found to have a stalk length of 265.5 μm as compared with 696.0 μm in *Dicyrtoma minuta*; the diameters of the droplets in these two species were 54.7 μm and 96.2 μm respectively. Droplets were retained at the tip of the stalk by a conical structure in *Dicyrtoma minuta* and a loop of the stalk in *Tomocerus minor*. Methods of sperm transfer are considered, and the possibility of the use in this connection of the ventral tube and ventral groove considered.

It may take from two minutes in *Isotoma sensibilis* to eighty minutes in *Dicyrtoma minuta* to lay a single egg. Mean egg batch sizes in mass culture vary from 4.6 in *Onychiurus procampatus* to 34.9 in *Isotoma olivacea*. Isolated females may lay more eggs, up to 51 being recorded in a single laying period of *Isotoma viridis*. In *Tullbergia krausbaueri* egg laying immediately follows moulting after the third and subsequent instars, and the number of eggs laid after each moult increases up to the seventh or eighth instar, and afterwards decreases. Estimates of the total number of eggs laid during life are made, and it is suggested that most Moor House species lay about 90 eggs during their lifetime of about one year.

Laying occurs mainly in spring, but in some species it may continue through the summer and into autumn. Egg development is described and developmental periods at different temperatures are recorded. A linear relationship between temperature and the reciprocal of the development time is demonstrated and developmental zeros are shown to range between 0 °C and 3 °C. Using calculated thermal constants estimates are made of the developmental times in the field.

8. Zusammenfassung

Das Vermehrungsverhalten einiger Collembolenarten aus dem Moor-House-National-Naturschutzgebiet im Westmorland/England (ca. 560 m ü. M.) wird beschrieben. Die Schlußfolgerungen von MAYER (1957), daß Poduriden und Onychiuriden keine Spermatophoren erzeugen, wird bestätigt. Es wurde gefunden, daß die Stiellänge bei Spermatophoren von *Tomocerus minor* 265,5 und vergleichsweise von *Dicyrtoma minuta* 690,2 μm beträgt. Die Durchmesser der Spermatophorenköpfchen dieser beiden Arten beträgt 54,7 bzw. 96,2 μm. Die Köpfchen werden bei *Dicyrtoma minuta* durch eine konische Struktur an der Spitze des Stieles festgehalten und bei *Tomocerus minor* durch eine Schlinge des Stieles. Methoden des Spermientransportes und in Verbindung damit die Möglichkeit des Gebrauchs des Ventraltubus und der Ventralgrube, werden in Betracht gezogen.

Es mag etwa 2 min bei *Isotoma sensilitis* und bis zu 8 min bei *Dicyrtoma minuta* dauern, bis ein einziges Ei gelegt wird. Die durchschnittliche Größe der Gelege variiert in Massenkulturen von 4,6 bei *Onychiurus procampatus* bis 34,9 Eier bei *Isotoma olivacea*. Isolierte Weibchen legen möglicherweise mehr Eier; bis zu 51 Stück sind bei *Isotoma viridis* beobachtet worden. Bei *Tullbergia krausbaueri* folgt die Eiablage unmittelbar auf die Häutung nach dem 3. und den folgenden Häutungsstadien. Die Anzahl der Eier, welche nach jeder Häutung gelegt werden, steigt vom 7. zum 8. Häutungsstadium an und nimmt dann wieder ab. Schätzungen der totalen Anzahl von Eiern, die während des Lebens eines Weibchens abgelegt wurden, werden vorgenommen. Es wird vermutet, daß die meisten Collembolenarten von Moor House etwa 90 Eier während ihrer rund einjährigen Lebensdauer legen.

Eiablagen erfolgen hauptsächlich im Frühling, aber bei einigen Arten mögen sie auch durch den Sommer bis in den Herbst hinein fortgesetzt werden. Die Eientwicklung wird beschrieben und über die Entwicklungsperioden bei verschiedenen Temperaturen wird berichtet. Eine lineare Beziehung zwischen Temperaturen und dem Kehrwert der Entwicklungsdauer wird nachgewiesen und es wird gezeigt, daß Entwicklungs-Nullpunkte zwischen 0 und 3 °C liegen. Mit Hilfe berechneter thermaler Konstanten werden die Entwicklungszeiten im Freiland geschätzt.

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